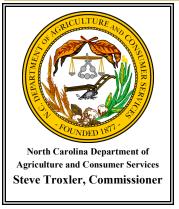




Laboratory Diagnosticians

Veterinary News and Information From North Carolina's Diagnostic Laboratories



NCVDLS-Rollins Lab 1031 Mail Service Center Raleigh, NC 27699-1031 Phone: (919) 733-3986 Fax: (919) 733-0454 Website:

In This Issue...

Client Corner	2
Client Corner	2
Feature Article	2
Short Cuts	5
Departmental News	16
Directory	17
Directory	

Holiday Closings...

September 5, 2016

November 11, 24 and 25, 2016

December 23, 26 and 27, 2016

Our laboratories will be closed

Please e-mail

NCVDL@ncagr.gov with any
comments and/or suggestions
concerning The NCVDLS

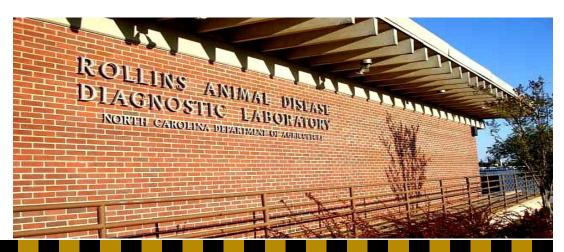
Report

Editor - Tamara Seago

Message from the Director

Since the release of the last newsletter it has been a time of transition at the NCVDLS. In October of 2015 Dr. Karen Post, the former laboratory director, retired. I hope you will join me in thanking Dr. Post for all her hard work, contributions, and service to the laboratory. I arrived at the Rollins laboratory in June of 2014 as the Pathology Services Coordinator. In October of 2015 I took over as Interim Director and as of February 2016 have officially been named the Lab Director. I could not be happier to be back in North Carolina to raise my family, and I am grateful for the opportunity provide veterinary diagnostics to the animal industry, veterinarians, and citizens of North Carolina. I feel fortunate to be able to work daily with our highly trained and dedicated laboratory team. My goal is to build on the solid foundation built by Dr. Post, as well as the previous Director's, in assuring quality remains paramount while we set about fulling our goal of providing accurate and timely diagnostic services.

Jim Trybus, DVM, DACVP



Feature Article

Blister beetle (cantharidin) toxicity

Clinical history

On the morning of September 2015, the Rollins Laboratory received a phone call from a veterinarian who had been called to examine 2 sick horses. A mare and pony were found in their stalls sweating and salivating, they felt hypothermic, and they were mildly ataxic. After they were led out of their stalls, they collapsed within a few feet and died a few minutes later. Three more horses on the farm had foul smelling diarrhea and decreased intestinal sounds that morning. The veterinarian discovered that the only common factor between the deceased horses and the three sick ones was the new bales of alfalfa hay that had been purchased earlier that week and fed to all affected horses. The three sick horses had only nibbled at the hay, so the hay had been removed and then fed to the 2 horses who ate all of the hay and died the next morning. Given that the new alfalfa hay was the common link with these animals the veterinarian was concerned about blister beetle toxicity. The pony and a flake of suspect alfalfa hay were submitted to the laboratory for necropsy. The hay was shaken and several striped beetles, consistent with the Striped Blister beetle, fell from the hay. Two more horses from this farm, who had also eaten the same alfalfa hay, died later in the day. The 3 horses who had only nibbled the hay were treated symptomatically and recovered.

Gross and histopathological lesions

The pony was necropsied and the subtle gross lesions observed included dark red to purple gastric mucosa and watery and red small intestinal contents. No oral ulcers were present in the pony's mouth. Based on these non-specific subtle lesions blister beetle toxicity may not have been considered in the differential diagnosis without the clinical history. Histologically there was acute congestion and petechiation in the lung, heart, kidney, and adrenal gland. Additionally, there was acute renal tubular degeneration, acute erosive enteritis, and erosive fibrinosuppurative gastritis. The histopathological lesions are supportive of a caustic agent targeting the GI and renal systems, however histopathology takes time to process the samples, which would have delayed a diagnosis and led to more deaths *if the referring veterinarian had not done an on-farm investigation*.



Striped blister beetles collected from hay at necropsy. (photo credit: Dr. Mahogany Wade-Caesar)



Stomach mucosa with congestion, erosions, and adhered fibrinosuppurative material.

Toxic principal

There are 2500 species of blister beetles and about 12 species are found in the central and/or southeastern U.S. Blister beetles vary by species in shape, size (3/8 to 1 inch long) and color (solid gray to black or with paler wing margins, metallic, yellowish striped or spotted). Most are long, cylindrical, narrow-bodied beetles that have heads that are wider than the first thoracic segment. The wing covers are usually soft and pliable. The name of these beetles comes from the substance, cantharidin, which is found in their hemolymph. This substance is a vesicant and irritant to the GI tract and urinary system. Cantharidin is even used in human medicine as the active ingredient in a proprietary wart remover. Taken internally or absorbed through the skin, cantharidin is highly toxic to mammals. The lethal dose for horses is estimated to be between 0.5 and 1.0 mg of cantharidin per kilogram (2.2 lbs) of body weight. The different species of blister beetle contain varying levels of cantharidin and the striped blister beetle contains the highest average concentration (5.0 mg/beetle) of the toxic substance. So 30-50 striped blister beetles may be fatal to an adult horse.

Cantharidin (mg / beetle)	250	Horse 500	Weight (lbs) 800	1200
20 000	Number	of	Blister	Beetles
0.5	227	450	719	1090
5.0	23	45	72	109

Table 1. Estimated lethal numbers of blister beetles of two cantharidin contents that must be ingested by horses of different weights. (https://entomology.ca.uky.edu/ef102)

These 4 blister beetles can be found in North Carolina.



Striped Blister beetle (Picture Credit: John L. Capinera, University of Florida; University of Florida IFAS Extension https://edis.ifas.ufl.edu/in323)



Black blister beetle (Picture Credit: John L. Capinera, University of Florida; University of Florida IFAS Extension https://edis.ifas.ufl.edu/in323)



Marginated blister beetle (Picture Credit: John L. Capinera, University of Florida; University of Florida IFAS Extension https://edis.ifas.ufl.edu/in323)



Ashgrav blister beetle (Picture Credit: John L. Capinera, University of Florida; University of Florida IFAS Extension https://edis.ifas.ufl.edu/in323)



Effects on livestock

Cantharidin is very stable and remains toxic in dead beetles for a long time. Animals may be poisoned by eating crushed beetles in cured hay. Ruminants can also be poisoned but horses are more sensitive. The severity of the clinical signs depends on the amount eaten. Clinical signs usually appear within hours and include depression, mild to severe colic, playing in the water with lips and tongue, straining during frequent urination (after 24 hrs urine may become dark from blood), fever, low serum calcium and magnesium, which may cause muscle stiffness or an exaggerated goose-stepping gait, or exaggerated contraction of the diaphragm muscle (diaphragmatic flutter). The calcium level can be drastically low and prolonged, which may damage the heart muscle. Laboratory derangements are usually hypocalcemia, hypomagnesemia, and azotemia. In acutely affected horses urine specific gravity is often less than 1.101 with hematuria with or without myoglobinuria. Death can occur within 6 to 72 hours.

Cantharadin toxicosis can be confirmed using high pressure liquid chromatography (HPLC) to detect and quantify cantharidin in the urine of live or dead horses, and in the gastric contents, liver, or kidneys of dead horses. At necropsy, erosions in the oral cavity, esophagus and stomach may be seen, as well as ulcerated to pseudomembranous enteritis. The most commonly reported gross pathologic lesions include necrosis and ulceration of the squamous lining of the distal esophagus, (fore)stomach and urinary bladder.

Beetles in alfalfa hay

Blister beetle species feed on flowers and foliage of a wide variety of crops including alfalfa, ornamental plants, potatoes, soybeans, garden vegetables and other plants.

Most blister beetle species have one generation per year. Females lay eggs in soil from late summer into early fall. Eggs are laid in clusters and hatch in about two weeks. The larvae move over the soil and find grasshopper eggs on which they feed and this is why outbreaks of blister beetles appear to follow grasshopper outbreaks. These larvae overwinter in the soil, pupate the following spring, and depending on the species, emerge from the soil in June, July, or August. Adults then feed, mate, and lay eggs for the next generation.

Blister beetle problems traditionally have been associated with alfalfa from arid, western states where environmental conditions encourage frequent grasshopper outbreaks. Although not as common, alfalfa from more eastern states may experience blister beetle problems in years following heavy grasshopper infestations.

Few if any blister beetles are present in the first cutting of alfalfa, but the cutting (usually second) that includes the flowers may be heavily contaminated with blister beetles who are attracted to flowering vegetation. These insects are very mobile and congregate in large numbers in small areas of the field. This habit makes them easier to locate in an alfalfa field, but also increases the chance of harvesting large numbers with the hay. Activity of the adult beetle ceases in early fall.

Blister beetle contamination in the hay is usually the result of alfalfa being crimped when cut so the beetles are crushed and are trapped in the hay. Beetles are also killed and trapped when the alfalfa is driven on before beetles have a chance to fly away. Cutting without using crimpers and avoiding driving over freshly cut alfalfa are two of the best ways to avoid heavy contamination of blister beetles.

There are 2 pesticides that are labeled for blister beetles with a 7 day pre-harvest waiting period.

Avoiding blister beetle toxicity (from Oklahoma Cooperative Extension Service PPD-2072: http://pods.dasnr.okstate.edu/docushare/dsweb/Get/Document-2080/EPP-2072.pdf)

Know your alfalfa supplier.

Ask producers what precautions were taken to avoid the presence of blister beetles in forage.

Inspect hay before feeding if the presence of blister beetles is suspected, although the lack of visible beetles does not completely eliminate the risk of cantharidin being present.

Purchase hay harvested before May or after September. This will not guarantee a lack of problems with blister beetles but can reduce the risks significantly.

If symptoms appear, call your veterinarian immediately.

Resources:

http://entnemdept.ufl.edu/creatures/urban/medical/blister beetles.htm

http://texasinsects.tamu.edu/bimg167.html

http://extension.missouri.edu/p/G4569

https://entomology.ca.uky.edu/ef102

https://www.addl.purdue.edu/newsletters/2006/Fall/EquineCT.htm

Short Cuts

COMPANION ANIMAL

Feline

A 12-week-old female Singapura kitten had a history of upper respiratory congestion, decreased activity, and exercise intolerance that progressed to coughing, choking, and snoring. Thoracic radiographs were unremarkable. The kitten was diagnosed with bilateral nasopharyngeal polyps. The masses were surgically debulked and the kitten died the following day. On necropsy, both polyps extended into the corresponding auditory tube and tympanic bulla. The lungs were semi-firm with diffuse congestion and edema. Histopathologic examination confirmed the nasopharyngeal polyps and revealed upper and lower respiratory infections, characterized by fibrinosuppurative rhinitis and bronchopneumonia with necrosis and edema.

A 7-month-old male domestic shorthair kitten was presented for a 2 month duration of upper respiratory infection, poor growth, and intermittent anorexia. Physical examination revealed poor body condition, copious ocular and nasal discharge, and sneezing. The kitten failed to respond to symptomatic treatment and was euthanized. On necropsy examination, the right and left nasal cavity was filled with copious amounts of mucopurulent material and there was moderate diffuse reddening of the nasal turbinate epithelium. The nasopharyngeal region was obstructed by a $3.5 \times 2 \times 1$ cm severely edematous tan smooth mass with multifocal hyperemia. A $7 \times 4 \times 2$ mm tan mass was identified in the right nasopharynx. The stalk of each mass extended into the corresponding auditory tube and tympanic bulla. Nasopharyngeal polyps and fibrinosuppurative rhinitis with necrosis and edema were identified via histopathology.

Nasopharyngeal polyps are noncancerous masses that originate from the middle ear or auditory tube epithelium. Blockage of the nasopharyngeal cavity may occur as the polyps enlarge and expand. Clinical signs may include nasal discharge, sneezing, stertor, dyspnea, dysphagia, gagging, voice change, head shaking, pawing at ears, vestibular signs, ataxia, Horner's syndrome, and/or facial nerve paralysis. The exact etiology is unknown, but a response to chronic inflammation (ie. respiratory viruses) has been proposed. Viral agents were not detected via immunohistochemistry or isolated via viral isolation in either felid. Nasopharyngeal polyps tend to occur in young cats, as seen in these two cases. Despite surgical debulking, the younger kitten died from bronchopneumonia and complications secondary to the polyps.



Figure 1. A 7-month-old DSH kitten with a left nasopharyngeal polyp and abundant mucopurulent nasal discharge.

COMPANION ANIMAL, CONTINUED

Equine

An 18 year old Thoroughbred mare had presented to the veterinarian for colic and decreased appetite. Blood work showed an increased Gamma-glutamyl transferase (GGT), Sorbitol Dehydrogenase (SDH), Bile Acids and Ammonia levels. A liver biopsy showed a biliary obstruction pattern with bridging fibrosis. The horse was euthanized due to the poor prognosis. On post mortem examination the horse weighed ~ 545 to 590 kg and was in good body condition. The liver was diffusely swollen, misshapen, firm on palpation with areas of fibrosis. There was marked dilatation of the bile ducts with material that ranged from thick, granular green material to a yellow mucoid liquid (Figure 1). A 6 cm by 5 cm by 4 cm cholelith was lodged in the common bile duct (Figure 2). There was a 28 cm by 30 cm by 4 cm thick area of thickened gastric wall within the glandular mucosa of the stomach. Multiple areas of gastric ulceration were present. The gastric wall thickening accompanied a ~ 5 cm wide mucosal ulcer with an underlying submucosal abscess (Figure 3). On histopathology, there was moderate to severe portal and bridging fibrosis with cholestasis, peribiliary fibrosis and mild lymphoplasmacytic inflammation of the liver. Examination of the stomach sections showed ulcerative gastritis with granulation tissue and hemorrhage.

The changes in the liver were considered the result of chronic cholestasis due to biliary obstruction. The cause of the gastric ulcer was not apparent.



Figure 1 – Liver cross section. Irregularly contoured surface with pale tan to white connective tissue replacing the normal parenchyma multiple, and multiple dilated bile ducts



Figure 2- Cholelith



Figure 3- Gastric ulcer with submucosal abscess

Volume 11 Issue 1 T

The NCVDLS Report

Page 9

LIVESTOCK

Cattle

Difficulty breathing quickly progressing to recumbence was the main sign observed recently in an 8 year old cow. A veterinary examination of the cow in the field did not identify a specific diagnosis and the cow was euthanized and submitted to Griffin Animal Disease Diagnostic Laboratory in Monroe, NC. Another cow in the same herd with a similar illness had been euthanized and disposed of on the farm about 10 days previous to the illness of this cow. Significant lesions were not identified upon gross examination, thus the cause of clinical illness in this animal remained uncertain. The brain was submitted for Rabies testing and the result was positive. This diagnosis illustrates the varied way that Rabies infection may present clinically in cattle. A review of the NCVDLS accessions since the beginning of 2016 indicates 56 cattle have been tested for Rabies. A total of 4 of these cattle have been positive. Three of these were calves ranging in age from 1 to 5 months and one was this 8 year old cow. Three of these cattle were geographically located so they were examined at the closest laboratory which was the Griffin Laboratory in Monroe, NC.

Making a diagnosis of Rabies requires laboratory testing and should be pursued certainly when Rabies is suspected and also in some cases where another diagnosis in uncertain. Your veterinarian should be consulted for examination of sick cattle. When Rabies is suspect or considered, the appropriate method of euthanasia should be chosen to ensure the brain remains intact. The head can be removed for testing or the entire carcass may be submitted.

Cattle can be vaccinated against Rabies. Although vaccination is not routinely performed in most herds.

Please consult your veterinarian.

Dr. Reginald Ridenhour

Client Corner

NC Invests in the future of Agriculture

It is an exciting time at the laboratory as with the passage of the NC Bond Referendum we have begun preliminary planning regarding a new NCDA & CS Agricultural Sciences Center that will replace the aging Veterinary Diagnostic, Food and Drug, Structural Pest Control and Pesticides, Standards, and Motor Fuels laboratories. The average age of these labs is more that 40 years. The co-located location in concert with a new and modern design will promote the ability to invest in advanced modern technology and testing equipment, be more energy-efficient, optimize workflows and expand programs to be better positioned to meet the future testing needs of North Carolina citizens and industry.

NCVDLS Recognized as NAHLN Tier 1 Laboratory

The Rollins laboratory was one of the original 12 core laboratories of the National Animal Health Laboratory Network (NAHLN) system which comprises a group of state and federal laboratories capable of performing surveillance testing for high-consequence agricultural and zoonotic diseases. Recently, NAHLN restructured its' network to meet overall national needs. Criteria considered during the restructure included: lab accreditation status, geographic location, population density of animals, testing capabilities, facilities and infrastructure status, farm gate values, and risk of foreign animal disease introduction. The Rollins laboratory has been recognized as a Level 1 laboratory, one of only 11 NAHLN labs. This is a credit to our capabilities, quality system, LIMS reporting, and testing expertise.

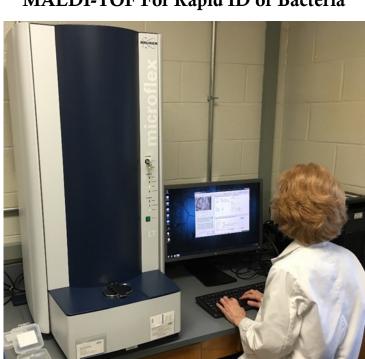
Client Survey:

There are plans to conduct another client survey this year so we can better assess your needs. If you receive a survey, please take the time to complete it and return it to us. We are continually striving to make improvement to better serve you and the survey will greatly assist us in doing so.

Service Fee for Avian Flu PCR Test:

On February 16th the NC Board of Agriculture approved a new fee for Avian Influenza PCR test. The \$6 per test fee is to be paid by the commercial poultry industry and will help our laboratory meet the growing demand for this test. The test will continue to be free for owners of backyard and hobby flocks. The charge became effective May 1, 2016.

Client Corner



MALDI-TOF For Rapid ID of Bacteria

NCVDLS invests in state-of-the-art technology that will improve laboratory efficiency and accuracy. The bacteriology section at Rollins is in the preliminary stages of transitioning bacterial identification to the Matrix-Assisted Laser Desorption Ionization-Time of Flight Mass Spectrometer, also known as the MALDI-TOF. The MALDI-TOF identifies bacteria and fungi based on the organism's protein composition. Once a microorganism is isolated it can be accurately identified within minutes with this technology rather than the 2-7 days currently required when using traditional identification methods.

ROLLINS LABORATORY

Meet the Staff: Receiving

Our receiving department gets a first-hand look at most of the cases arriving at our testing facilities. They are responsible for opening submissions, evaluating the samples submitted and ensuring these match the test(s) requested. Also initial entry of the submission information into our LIMS, and delivery of the specimens to appropriate laboratory sections for testing occurs in this department. They receive up to 300 submissions a day and never quite know what to expect when they are opening packages.



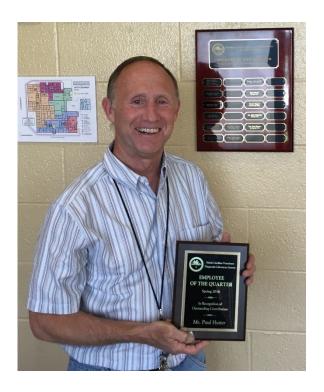
Becky Boone, seen here entering case/client information into our laboratory reporting system, has been with the receiving department for four years.



Dell Weaver (front) and Dominic Gonzalez (back) are busy checking the samples with the corresponding submission information. Dell has been with the receiving department for twenty two years and Dominic, who lends a hand to several laboratory sections, started in fall of 2015.

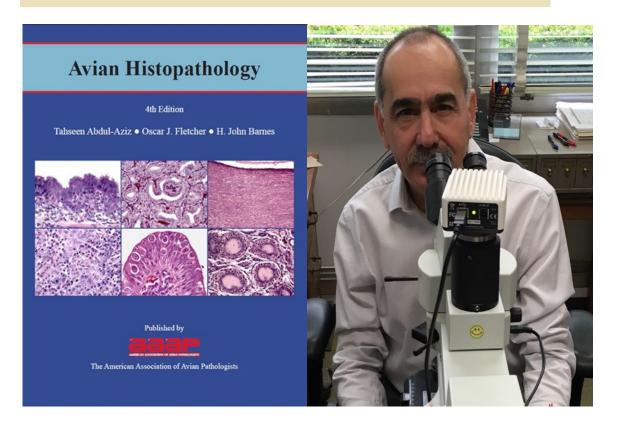
ROLLINS LABORATORY

Employee of the Quarter



I would like to congratulate Mr. Paul Hutter for being named Employee of the Quarter. Paul is our safety officer. He often interjects humor with his daily responsibilities of maintaining a safe working environment for our laboratory team. Paul volunteered to oversee our weekly laboratory meetings regarding Avian Influenza preparedness that enabled us to identify and address efficiencies, with the help of many employees in several laboratory sections, regarding biosecurity, sample receiving, sample processing, and testing. Paul even developed and administered a fun and interactive table-top exercise simulating an avian influenza outbreak scenario that allowed us to work through our responses to a disease event.

ROLLINS LABORATORY



Tahseen Aziz, DVM, MS, PhD, Diplomate ACPV is employed by the North Carolina Veterinary Diagnostic Laboratory as our Avian Pathologist. Dr. Aziz has been integrally involved in the completion of the 4th Edition of the Avian Histopathology. The new edition has about 650 pages that include 2005 colored pictures. Several chapters in this edition have been revised by incorporating new information and adding new images. As in previous editions, the book maintains the organization of chapters by organ systems, which provides a systemic approach to avian histopathology. Lesions associated with specific diseases and conditions in different organs and tissues are described. The 4th edition of the Avian Histopathology will be available for purchase in June 2016.

Congratulations to Dr. Aziz for the foresight, hard work, and dedication that was required in completing this project!

New Hires

Dominic Gonzalez - 9/21/15 Carol Garbarino - 12/21/15 Jennifer Burgoyne - 1/06/16 Jenna Hunter - 2/29/16 Dr. David Ackerman - 3/01/16 Dana Camp - 3/17/16 Marcia Sutton - 4/18/16

Retirements

Dr. Karen Post - 12/01/15 Helen Tang - 12/01/15 Fuhing Lo - 3/01/16

Service Award Recipients

Kim Gooden - 30 yrs
Paul Miller - 30 yrs
Kathy Harwood - 25 yrs
Kristie Long - 20 yrs
Dr. Stacy Robinson - 15 yrs
Dr. Mary Swanson - 10 yrs
Dr. Tahseen Aziz - 10 yrs
Cindy Orlosky - 10 yrs

Directory

Rollins Laboratory - 919-733-3986

Director

Dr. James Trybus

Assistant Director

Dr. Richard Mock

Veterinary Pathologists

Dr. Tahseen Abdul-Aziz (Avian)

Dr. Allison Boone (Anatomic)

Dr. Steven Rushton (Anatomic)

Dr. Alison Tucker (Anatomic)

Veterinary Diagnosticians

Dr. Jennifer Haugland

Dr. Stacy Robinson

Dr. Mahogany Caesar

Microbiologists

Dr. Richard Mock

Dr. Neeti Dahal

Laboratory Section Supervisors

Younghee Lee—Virology

Sandy Murphy—Bacteriology

Mary Baker—Histopathology

Dr. Kristen Crook—Serology

Beverly Wood—Molecular Diagnostics

Quality System Manager

Tina Buffington

Ghazala Jawad

Western Laboratory

785 Airport Rd.

Arden, NC 28704 Phone: (828) 684-8188

Fax: (828) 687-3574

Northwestern

Laboratory

1689 N Bridge St. Elkin, NC 28621

Phone: (336) 526-2499

Fax: (336) 526-2603

Griffin Laboratory

401 Quarry Rd. Monroe, NC 28111

Dr. Allen Cannedy

Mr. Larry Wooten

Phone: (704) 289-6448

Fax: (704) 283-9660

Director

Dr. Richard Oliver

Veterinary Diagnostician

Dr. David Drum

Director

Dr. David Ackerman

Veterinary Diagnostician

Dr. Jessica Kees

Director

Dr. Kim Hagans

Veterinary Diagnostician

Dr. Reg Ridenhour

Diagnostic Laboratory Advisory Committee

Small Ruminant/Camelid Practitioner

· ·	
Dr. Eric Gonder	Corporate Poultry Practitioner—Goldsboro Milling
Dr. Jennifer Haugland	Veterinary Diagnostician—NCDA&CS Veterinary Diag-
Dr. Shannon Jennings	nostic Laboratory System Corporate Poultry Practitioner—Nash Johnson Farms
Dr. Randy Jones	Private Veterinary Practitioner—Livestock Veterinary Services
Dr. Richard Kirkman	Private Veterinary Practitioner—Large Animal
Dr. David Marshall	State Veterinarian—NCDA&CS Veterinary Division
Dr. James Trybus	Director of Laboratories—NCDA&CS Veterinary Diagnostic Laboratory System
Dr. Rick Sharpton	Corporate Poultry Practitioner—Perdue, Inc.
Dr. Betsy Sigmon	Small Animal Practitioner—Creature Comforts Animal Hospital

N.C. Farm Bureau